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STANISŁAWA CIANCIARA

BIOMETRIC AND BIOENERGETIC CHARACTERIZATION OF THE DEVELOPMENT OF *CLOËON DIPTERUM* (L.)

Department of Ecological Bioenergetics, Institute of Ecology, Polish Academy of Sciences, Dziekanów Leśny, 05-150 Łomianki, Poland

ABSTRACT

Biometric measurements (1149 determinations of body length, head capsule breadth, and wet and dry weight), as well as determinations of the caloric value (89 combustions) and oxygen consumption at 20°C (232 measurements) were performed in the aquatic stages (larvae and nymphs), and winged stages (subimago and imago) of *Cloëon dipterum*. It was found that in this species the linear body dimensions increase in proportion to the number of the stage, and that the curves of the increase in wet and dry body weight are S-shaped. The development of the aquatic forms of *C. dipterum* was characterized by a constant rate of the increase in dry weight relative to body length and by a constant increase in the caloric value of the body. In the development two periods differing in the intensity of the physiological processes were distinguished: period I (stages L_{I}—L_{V}) characterized by a more rapid rate of the increase in wet weight and by a rise of body hydration; period II (stages L_{VI}—N) characterized by a higher oxygen consumption rate and by a drop in body hydration.

1. INTRODUCTION

Cloëon dipterum, being a typically eurybiontic species (Fox, Simmonds 1933; Fox et al. 1937; Whitney 1939; Wingfield 1939; Dorier, Vaillant 1954; Chaurin 1956; Kamler 1971; Brittain 1974), also exhibits a wide range of geographic distribution. In the Paleoarctic it occurs widely in the lenitic waters of the whole Europe (Illies 1967), in many environments being the predominant species among Ephemeroptera (Harnisch 1957; Sowa 1959; Ikonomov 1960; Macan 1962; Kamler 1971; Keffermüller 1972). Larvae and nymphs of *C. dipterum*, often occuring in masses, can be of great importance for the biocenoses which they inhabit and may be an essential link of the trophic chain.

In ecological studies of a population it is indispensable to know the values of the biometric and bioenergetic parameters as well as to know their interrelations in the development of the investigated species. Although *C. dipterum* being a widely spread species is often the object of various studies, the relevant literature data are rather fragmentary; namely, they usually concern only one selected parameter and they report the mean values for the total development. Objectives of the present study were to investigate the course of changes in body proportions, hydration and caloric value of the body as well as in the oxygen consumption; moreover, the aim was to determine the correlations between body dimensions, body weights and maintenance costs during the development of *C. dipterum*.

2. MATERIAL AND METHODS

Experimental material

Cloëon dipterum (Linné) of genus *Cloëon* (Leach 1815) (Ephemeroptera : Baetidae) was determined with the use of the keys by Mikulski (1936), Bogoescu (1958), and Sowa (1975).

The material of larvae and nymphs of *C. dipterum* originated from an isolated small water body (in Wawer, near Warsaw). The specimens caught were, together with water, algae and mud from the natural environment, transferred to the laboratory where after 1—2 weeks' acclimatization at 293 K (20°C) they were used for respiration measurements (also at 20°C). The material employed for the biometric measurements and combustions was preserved in a 4% formalin solution.

Biometric parameters (body dimensions and body weights) and the bioenergetic parameters (caloric value, oxygen consumption) were determined for the aquatic stages — larvae and nymphs, and for the winged forms — subimago and imago; the dimensions of the mesothoracic wing pads relative to the successive segments of the thorax and abdomen of the larva served as a criterion for classification into stages (Cianciara 1979a). All investigated individuals belonged to the winter generation of *C. dipterum* (Cianciara 1979b).

Biometric measurements

For the investigated stages (larvae L_{I}—L_{VII}, nymphs, subimago and imago), measurements were made of: body length, head capsule breadth, and wet and dry body weight of each individual. A total of 1149 individuals of *C. dipterum* were measured.

Body length from the apex of the head to the base of the tail (l, mm), and the breadth of the head capsule (h, mm) were measured (Fig. 1) using a stereoscopic microscope (magn. 3×8) with a measuring eyepiece (one scale — 0.03 mm).

Wet body weight (W_{wt}, mg) was measured after drying of animals on filter paper, whereas the dry body weight (W_{dry}, mg) was determined upon drying of single individuals in aluminium foil vessels in a desiccator over NaOH, at 50°C, to constant weight (Dowgiałło 1975). Measurements performed using a Sartorius analytical balance were exact to 0.01 mg.

For each stage calculation was made of the mean values ($\bar{x}$), standard deviations (± SD), and confidence intervals (± ci) of the linear dimensions (l and h) and body weights (W_{wt} and W_{dry}). Results obtained for the successive stages were compared statistically. It was assumed that probability $p < 0.01$ indicates high significance (HS), $0.05 > p \geqslant 0.01$ — significance (S) and $p \geqslant 0.05$ — no significance (NS). If the differences were found to be significant (Bailey 1959), their numerical value was calculated: $\bar{x}_1 - \bar{x}_2 \pm L_{0.05}$ where $L_{0.05}$ is the 95% confidence interval of difference between means — Oktaba (1974).

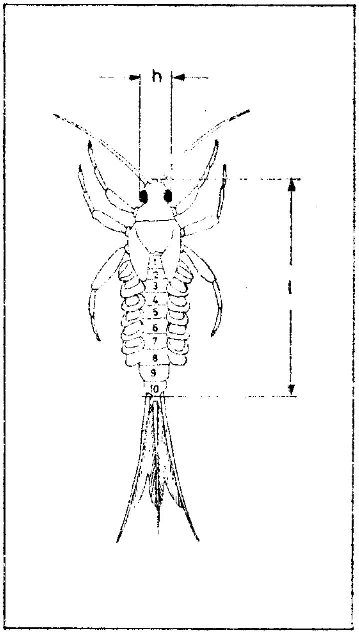

Fig. 1. Schema of biometric measurements of *C. dipterum*: l — body length, h — head capsule breadth; 1—10 abdominal segments

The percentile content of dry weight against body weight of a mean individual was calculated for each stage. The relationship between wet weight (W_{wt}, mg) and dry weight (W_{dry}, mg) was described by a linear regression equation: $W_{wt}=a+b\cdot W_{dry}$ (Pattée 1968). The relationships between wet weight (W_{wt}, mg) and dry weight (W_{dry}, mg), and the linear body dimensions (length — l, mm and head capsule breadth — h, mm) were described by logarithmic regression equations. By using the significance tests for comparison of the coefficients of two regressions (Bailey 1959, Simpson et al. 1960), the results obtained for the successive stages were analyzed. For the groups of stages, for which the differences in the regression coefficients between the different stages were insignificant (NS), moreover the common regressions for groups were calculated.

Determination of caloricity

The caloric value was determined directly by combustion in a adiabetic bomb microcalorimeter (Phillipson 1964), as modified by Klekowski (Klekowski, Bęczkowski 1973).

The caloric value (cal/mg dry weight) was measured for seven larval stages, nymphs, subimago and imago of both sexes of *C. dipterum*. For each stage, the body caloricity was determined in 6—10 replicates. A total of 89 combustions were carried out. Moreover, 5 determinations of the caloric value of the exuviae were performed.

From so determined the caloric value and mean dry body weights of individuals, the caloric equivalent of the body (E, cal/indiv.) was calculated for each stage. For conversion to the SI system units: 1 cal = 4.184 J.

Oxygen consumption measurements

Oxygen consumption was determined at 20.0°C ± 0.015 (293 K). The youngest larval stages (L_I L_{II}, L_{III} — Cianciara 1979a) were studied in Cartesian divers by the procedure described by Klekowski (1971). In the measurements of the amount of oxygen consumed by *C. dipterum*, one young individual was placed in each Cartesian diver. A total of 23 respiration measurements in the divers were carried out.

Moreover, 209 measurements of oxygen consumption (all stages) were performed in volumetric microrespirometers, as modified by Klekowski (1975). In the respirometric vessels the investigated individuals (either 2—5 indiv. of stages L_I—L_V or 1 indiv. of the older stages per vessel) were placed in 0.3 cm^3 of water. As CO_2 absorber, 0.05 cm^3 of 25% NaOH solution was used. Respirometers were equipped with capillaries (capacity about 10 mm^3) permitting readings exact to 0.05 mm^3 O_2.

Animals removed from the respirometers after the oxygen consumption measurements were preserved (4% formalin solution), measured (*l*, mm and *h*, mm) and weighed (W_{wt}, mg and W_{dry}, mg).

For each developmental stage of *C. dipterum* the mean volume of oxygen consumed (*R*, $mm^3\,O_2$/indiv.·h) was calculated; the values obtained for the successive stages were compared statistically. For some selected stages, the mean oxygen consumption rate ($mm^3\,O_2$/h·mg wet weight and $mm^3\,O_2$/·mg dry weight) was also calculated.

The oxycaloric coefficient was assumed to be 0.005 cal/$mm^3\,O_2$ (Winberg 1968; Trama 1957, has also used this value for mayfly *Stenonema pulchellum*). Multiplication of the oxygen consumption by this coefficient gave the caloricity losses for respiration.

The relationships between the oxygen consumption (*R*, $mm^3\,O_2$/indiv.·h) and body weight of animal (W_{wt}, mg and W_{dry}, mg) were described by logarithmic regression equations, and the relationships between the oxygen consumption and the linear dimensions (*l*, mm and *h*, mm) — by exponential regression equations. With the use of the significance tests for comparison of the exponents of two regressions (Bailey 1959; Simpson et al. 1960), the above-mentioned relationships were analyzed for the successive stages. For the developmental periods, for which no significant differences between the successive stages were found, one common regression was calculated.

3. RESULTS

A. BIOMETRY

Biometric measurements were perfomed in 1149 individuals belonging to all developmental stages of *C. dipterum*: larvae (L_I—L_{VII}), nymphs (N_I, N_{II}), subimago (S) and imago (A). For stages N_I, N_{II}, S and A, 113 females and 116 males were measured.

Body length (*l*, mm) and head capsule breadth (*h*, mm), found for the different developmental stages of *C. dipterum*, are presented in Fig. 2 and Tab. I. Although the ranges of the minimum and maximum values (Tab. I) for the successive stages overlap, significant (HS) differences in the mean values of the measured dimensions between stages from L_I to N_I were found; as concerns the nymphs and winged forms of both

Table I. Mean linear dimension (*l*—body length and *h*—head capsule breadth) in the succesive developmental stages of *Cloëon dipterum*

Stage		n	Body length (mm)			$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$ (or t)	p	Head capsule breadth (mm)			$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$ (or t)	p	l/h
			min	max	$\bar{x}\pm$SD			min	max	$\bar{x}\pm$SD			
L_{I}		70	1.98	2.97	2.55±0.26			0.24	0.51	0.38±0.08			6.64
						0.55±0.08	HS				0.12±0.02	HS	
L_{II}		198	2.64	3.75	3.10±0.30			0.36	0.60	0.50±0.08			6.19
						0.57±0.07	HS				0.11±0.02	HS	
L_{III}		188	3.21	4.29	3.67±0.38			0.45	0.72	0.61±0.08			5.99
						0.60±0.10	HS				0.12±0.02	HS	
L_{IV}		127	3.63	5.34	4.27±0.52			0.60	0.90	0.73±0.09			5.82
						1.01±0.14	HS				0.15±0.02	HS	
L_{V}		131	4.32	6.50	5.28±0.60			0.72	1.05	0.88±0.10			6.00
						1.13±0 16	HS				0.15±0.03	HS	
L_{VI}		106	5.37	7.26	6.41±0.62			0.90	1.20	1.03±0.09			6.21
						0.96±0.19	HS				0.14±0.03	HS	
L_{VII}		102	6.00	8.43	7.37±0.76			1.02	1.32	1.17±0.09			6.32
						1.39±0.22	HS				0.22±0.03	HS	
Females	N_{I}	55	7.47	9.39	8.76±0.45			1.23	1.50	1.39±0.10			6.32
						0.36±0.21	HS				$t=1.30$	NS	
	N_{II}	28	8.40	10.29	9.12±0.45			1.29	1.50	1.42±0.11			6.43
						$t=0.37$	NS				0.08±0.06	S	
	S	21	7.80	9.90	9.19±0.79			1.41	1.59	1.50±0.06			6.13
						$t=0.32$	NS				$t=1.78$	NS	
	A	9	8.34	10.08	9.29±0.90			1.29	1.50	1.45±0.07			6.40
						0.35±0.21	HS				0.18±0.03	HS	
Males	N_{I}	63	6.93	8.85	7.72±0.49			1.14	1.50	1.35±0.11			5.72
						0.40±0.22	HS				$t=1.46$	NS	
	N_{II}	30	7.38	9.03	8.12±0.54			1.26	1.47	1.38±0.07			5.88
						$t=0.46$	NS				0.10±0.05	S	
	S	16	7.47	9.75	8.21±0.80			1.38	1.59	1.48±0.08			5.56
						$t=0.65$	NS				$t=0.51$	NS	
	A	7	7.29	9.24	8.43±0.62			1.38	1.53	1.46±0.06			5.77

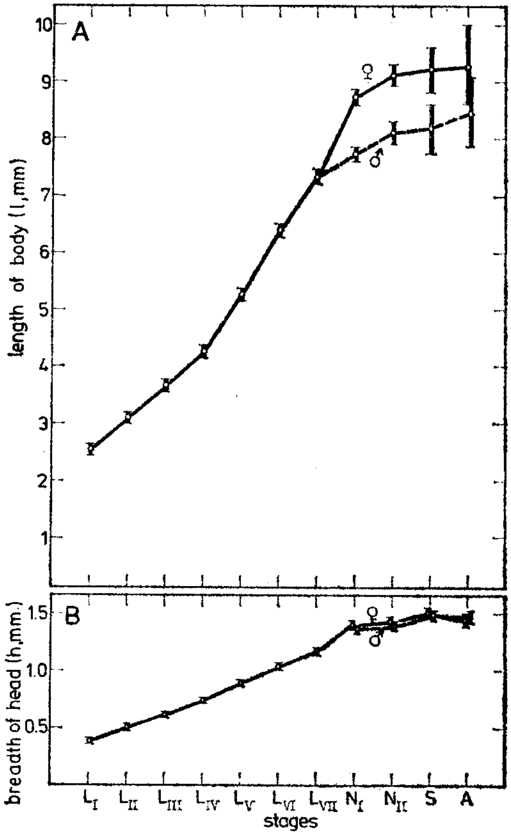

Fig 2. Linear body dimensions ($x \pm$ ci) in the successive developmental stages of *C. dipterum*: A — body length (l), B — head capsule breadth (h)

sexes, the linear dimensions either showed no differences (NS) or the differences were of low significance (S). Females (Tab. II, Fig. 2A) were much longer than males (HS) for all stages, except that of imago for which the differences in body length were at the limit of significance (S). This was doubtless due to the low number of individuals measured (9 females and 7 males); the winged forms were not, however, the main object of the present studies. On the other hand, there was no difference (NS) (Tab. II, Fig. 2B) in the mean head capsule breadth between females and males.

The curves of the increase in wet (W_{wt}, mg) and dry (W_{dry}, mg) weight of a mean individual in the development of *C. dipterum* were S-shaped (Fig. 3). Differences in body weights of mean individuals between the successive stages were significant (HS) (Tab. III), with the exception of the dry weight of the nymphs and winged forms of both sexes, for which the differences were NS or S. Females were much heavier (HS) than males (Tab. IV, Fig. 3).

Moreover, the weight of exuviae was measured for each stage. Dry weight of exuviae was expressed as percentage against the dry body weight. Larval exuviae accounted for about 5% of dry body weight of

Table II. Mean liner dimensions as a function of sex in *C. dipterum*

Stage	Body dimensions:	Body length (mm)		Head capsule breadth (mm)	
	Sex:	♀♀	♂♂	♀♀	♂♂
N_I	$\bar{x}\pm SD$	8.76±0.45	7.72±0.49	1.39±0.10	1.35±0.11
	t	11.65–HS		1.97–NS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	1.04±0.17		—	
N_{II}	$\bar{x}\pm SD$	9.12±0.45	8.12±0.54	1.42±0.11	1.38±0.07
	t	7.65–HS		1.64–NS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	1.00±0.27		—	
S	$\bar{x}\pm SD$	9.19±0.79	8.21±0.80	1.50±0.06	1.48±0.08
	t	3.80–HS		0.91–NS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	0.98±0.55		—	
A	$\bar{x}\pm SD$	9.29±0.90	8.43±0.62	1.45±0.07	1.46±0.06
	t	2.16–S		0.24–NS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	0.86±0.82		—	

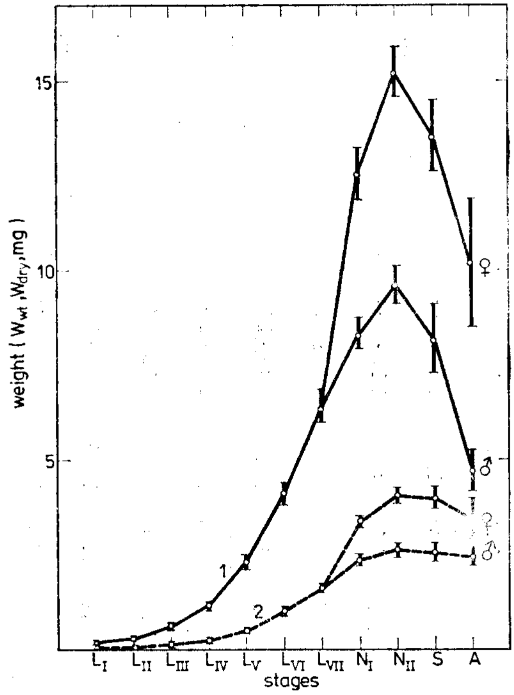

Fig. 3. Body weights ($\bar{x}$ ± ci) of a mean individual in the successive developmental stages of *C. dipterum*: 1 — wet weight (W_{wt}), 2 — dry weight (W_{dry})

Table III. Mean body weight (W_{wt}—wet weight and W_{dry}—dry weight) in the successive developmental stages of *Cloëon dipterum*

Stage		n	Wet weight (mg)			$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$ (or t)	p	Dry weight (mg)			$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$ (or t)	p	W_{dry}/W_{wt} (%)
			min	max	$\bar{x}\pm$SD			min	max	$\bar{x}\pm$SD			
L_I		70	0.05	0.31	0.17±0.07			0.01	0.09	0.04±0.02			23.66
						0.14±0.04	HS				0.04±0.01	HS	
L_{II}		198	0.09	0.64	0.31±0.14			0.03	0.18	0.08±0.04			24.42
						0.28±0.05	HS				0.06±0.01	HS	
L_{III}		188	0.25	1.42	0.59±0.28			0.05	0.30	0.14±0.07			23.63
						0.54±0.09	HS				0.12±0.07	HS	
L_{IV}		127	0.48	2.31	1.13±0.49			0.10	0.53	0.26±0.10			23.17
						1.16±0.17	HS				0.23±0.03	HS	
L_V		131	0.87	4.10	2.29±0.84			0.20	0.81	0.49±0.10			21.83
						1.81±0.26	HS				0.52±0.05	HS	
L_{VI}		106	1.92	6.92	4.10±1.21			0.55	1.74	1.01±0.28			24.70
						2.34±0.48	HS				0.60±0.12	HS	
L_{VII}		102	3.13	12.20	6.44±2.17			0.67	2.65	1.61±0.55			24.89
						6.10±0.82	HS				1.79±0.22	HS	
Females	N_I	55	8.61	17.84	12.54±2.91			1.96	4.90	3.37±0.84			26.85
						3.68±1.19	HS				0.68±0.35	HS	
	N_{II}	28	13.55	21.24	16.22±1.62			3.24	5.40	4.05±0.52			24.95
						−2.67±1.12	HS				t=0.38	HS	
	S	21	11.19	16.99	13.55±2.18			2.13	4.64	3.98±0.62			29.40
						−3.36±1.80	HS				−0.54±0.53	S	
	A	9	7.48	12.19	10.19±2.01			1.95	4.19	3.44±0.62			33.72
						1.90±0.65	HS				0.77±0.18	HS	
Males	N_I	63	5.31	12.54	8.34±1.78			0.92	3.69	2.38±0.56			28.53
						1.26±0.76	HS				0.24±0.23	S	
	N_{II}	30	8.10	11.37	9.60±1.50			1.80	3.77	2.62±0.48			27.27
						−1.42±1.01	HS				t=0.30	NS	
	S	16	5.70	11.58	8.18±1.71			1.73	3.19	2.57±0.55			31.47
						−3.46±1.44	HS				−0.62±0.48	S	
	A	7	3.67	5.40	4.72±0.59			1.60	2.36	1.95±0.29			41.38

Table IV. Mean body weights as a function of sex in *Cloëon dipterum*

Stage	Body weight: Sex:	Wet weight (mg) ♀♀	 ♂♂	Dry weight (mg) ♀♀	 ♂♂
N_I	$x\pm$SD	12.54±2.91	8.34±1.78	3.37±0.84	2.38±0.56
	t	9.28–HS		7.40–HS	
	$(x_1-\bar{x}_2)\pm L_{0.05}$	4.20±0.88		0.99±0.26	
N_{II}	$\bar{x}-$SD	16.22±1.62	9.60±1.50	4.05±0.52	2.62±0.48
	t	16.11–HS		10.97–HS	
	$(x_1-\bar{x}_2)\pm L_{0.05}$	6.62±0.84		1.43±0.27	
S	$x\pm$SD	13.55±2.18	8.18±1.71	3.98±0.62	2.57±0.55
	t	8.13–HS		7.18–HS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	5.37±1.38		1.41±0.41	
A	$x\pm$SD	10.19±2.01	4.72±0.59	3.44±0.62	1.95±0.29
	t	6.92–HS		5.80–HS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	5.47±1.80		1.49±0.58	

larvae (stages L_I to L_{VII}). Exuviae of nymphs (N_I and N_{II}) represented about 5 and 7.5%, respectively, of dry body weight of females and about 7 and 11.5%, respectively, of dry body weight of males. Subimaginal exuviae accounted for about 13 and 16% of dry body weight of females and males, respectively.

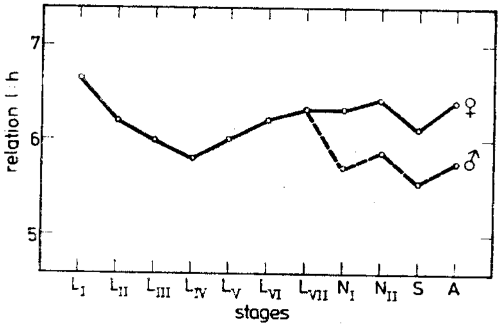

Fig. 4. Changes in the proportion of body length (*l*) to head capsule breadth (*h*) in the successive developmental stages of *C. dipterum*

In the larval development (Tab. I, Fig. 4) small changes in the body proportions, expressed by the ratio of body length to head capsule breadth were observed; on the average, for larvae: $l/h=6.17$. In stages for which sex was distinguished (nymphs and winged forms) the females were slimmer (on the average: l/h — 6.33) than males (on the average: $l/h=5.73$).

Hydration of the body of larvae (L_I—L_{VII}) was approximately constant; for these stages the mean content of dry weight (Tab. III, Fig. 5) was about 24%. Stages from L_I to L_V exhibited a small drop in dry weight content (from 23.7% to 21.8%), and then an increase to 33.7 and 41.4% in imago of females and males, respectively.

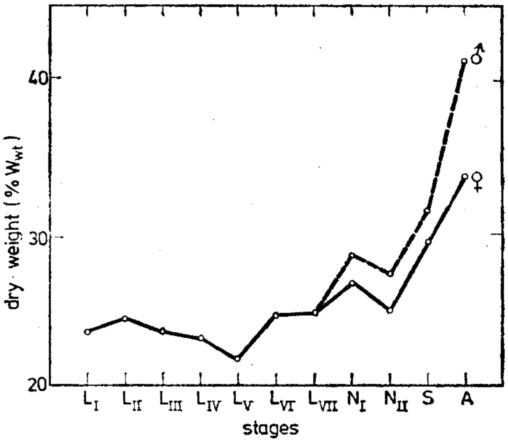

Fig. 5. Changes in percentile content of dry weight against body weight in the successive developmental stages of *C. dipterum*

The relationship between wet (W_{wt}, mg) and dry (W_{dry}, mg) weight was calculated for 1096 larvae and nymphs weighing from 0.05 mg to 21.24 mg wet weight. This relationship was described by the linear regression equation:

$$W_{wt}=0.16+3.95\ W_{dry}$$

where: correlation coefficient $r=0.97$ (95% confidence interval: for intercept $a=0.1583\pm0.0196$, and for slope $b=3.9547\pm0.1588$).

The relationships between wet (W_{wt}, mg) and dry (W_{dry}, mg) weight, and between body length (l, mm) and head capsule breadth (h, mm) were described by logarithmic regression equations (Tab. V). It was found that only the relationship between dry weight and body length (Fig. 6) during the development of the aquatic forms of *C. dipterum* remained relatively constant; the fact that the points on the plot, corresponding to individuals with dry weight below 0.06 mg, somewhat depart from the regression line may result from too great a weighing error (±0.01 mg), as compared with the small dry weight of individuals of the youngest stages. On the grounds of 1096 measurements of individuals with body length from 1.98 mm to 10.29 mm, belonging to stages L_I—N_{II}, this relationship was expressed by the equation:

$$W_{dry}=0.001\ l^{3.68}.$$

This equation, in logarithmic form and upon the application of 95% confidence limits for coefficients a and b, assumes the form:

$$\log W_{dry}=(3.0155\pm0.0342)+(3.6802\pm0.0489)\log l.$$

The high correlation coefficient ($r=0.98$) points to a close correlation between body length and dry weight of larvae and nymphs of *C. dipterum*.

Table V. Relationship between body weights (W_{wt}, mg and W_{dry}, mg) and linear body dimensions (l, mm and h, mm) in *Cloëon dipterum*

Relationship calculated	Stages	n	Regression equations (with 95% confidence interval for coefficients a and b)	r	Differences in coefficients a and b of regressions between two groups of stages
W_{wt} (mg) and l (mm)	L_I-L_V	712	$W_{wt}=(0.0041-0.0054)\cdot l^{3.5257-3.7353}$	0.9312	$t_a=$ 4.37 – HS
	L_{VI}-N	384	$W_{wt}=(0.0124-0.0236)\cdot l^{2.9493-3.2639}$	0.9019	$t_b=$ 3.90 – HS
W_{dry} (mg) and l (mm)	L_I-N	1096	$W_{dry}=(0.0009-0.0011)\cdot l^{3.6313-3.7291}$	0.9756	$t_a=$ 1.62 – NS $t_b=$ 1.04 – NS
W_{wt} (mg) and h (mm)	L_I-L_V	712	$W_{wt}=(2.4155-2.7227)\cdot h^{2.8249-3.0231}$	0.9089	$t_a=$11.99 – HS
	L_{VI}-N	384	$W_{wt}=(3.9120-4.3152)\cdot h^{2.6909-3.0985}$	0.8191	$t_b=$ 0.25 – NS
W_{dry} (mg) and h (mm)	L_I-L_V	712	$W_{dry}=(0.5580-0.6229)\cdot h^{2.8394-3.0220}$	0.9214	$t_a=$14.18 – HS
	L_{VI}-N	384	$W_{dry}=(0.9101-1.0025)\cdot h^{3.0782-3.4806}$	0.8537	$t_b=$ 3.10 – HS

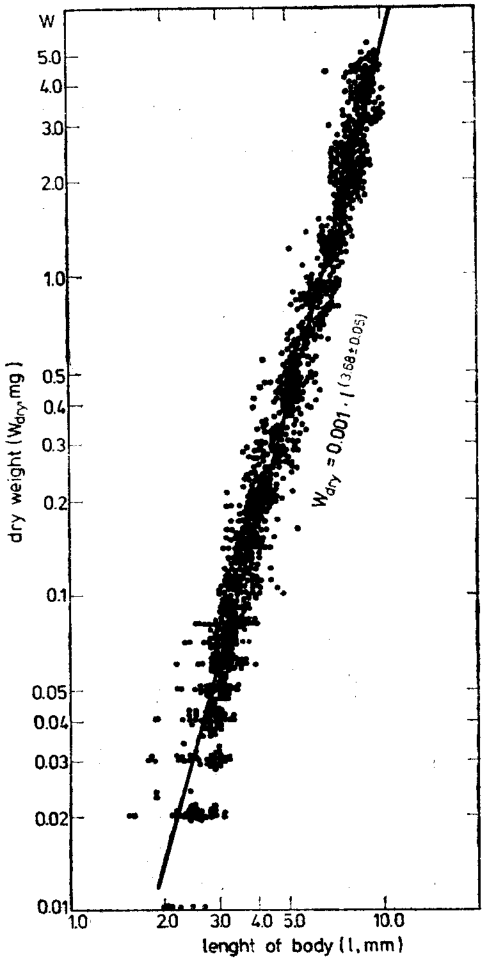

Fig. 6. Relationship between dry weight (W_{dry}, mg) and body length (l, mm) in *C. dipterum* (n=1096)

The remaining relationships (Tab. V, Fig. 7) were different. They did not remain constant during the development of the aquatic stages, but they differed for two groups of stages: from L_I to L_V and from L_{VI} to N_{II}. The differences between these groups were significant (HS).

For the winged forms (subimago and imago jointly) the relationship W_{dry}/l was calculated; it was found that:

— for females: $W_{dry}=0.144\ l^{1.51}$ (where $n=30$; correlation coefficient $r=0.74$; body length: 7.80—10.08 mm),

— for males: $W_{dry}=0.035\ l^{1.98}$ (where $n=23$; correlation coefficient $r=0.79$; body length: 7.29—10.79 mm). The differences between the coefficients of these regressions were insignificant (NS). On account of too narrow a range of the head capsule breadth for the winged stages, the equations of the relationships W_{wt}/h and W_{dry}/h could not be formulated.

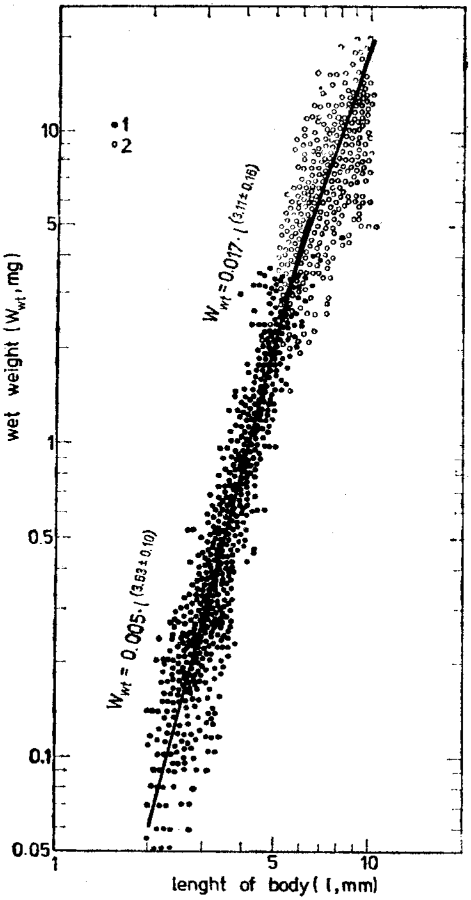

Fig. 7. Relationship between wet weight (W_{wt}, mg) and body length (l, mm) in *C. dipterum*: 1 — stages L_I—L_V (n=712), 2 — stages L_{VI}—N (n=384)

Mathematical description of the highly significant relationships between dry weight and body length, as well as between the wet and dry weight of *C. dipterum* may be of great importance for research, since it permits us to predict accurately — on the basis of the knowledge of one body parameter — the remaining ones.

B. CALORIC VALUE AND CALORIC EQUIVALENT

The caloric value of the body of *C. dipterum*, i.e. the caloricity calculated per unit of weight (cal/mg dry weight) increased with the development of larvae (Tab. VI); for stage L_{VII} it was about 20% higher than for stage L_I. During the development the maximal caloric value was found for subimago of both sexes, and a slightly lower one — for the imago stage.

Table VI. Caloric value and caloric equivalent (E) in some selected developmental stages of *Cloëon dipterum*; 1 cal=4.184 J

Stage		Caloric value cal/mg dry weight $\bar{x}\pm$SD	Caloric equivalent	
			cal/indiv.	cal/exuvia
L_I		5.0233±0.0743	0.2009	0.0108
L_{II}		5.2578±0.1480	0.3943	0.0203
L_{III}		5.5698±0.5302	0.7798	0.0379
L_{IV}		5.7092±0.3574	1.4958	0.0709
L_V		5.7168±0.3044	2.8012	0.1326
L_{VI}		6.0088±0.1914	6.0869	0.2741
L_{VII}		6.0716±0.4248	9.7692	0.4354
Females	N_I	6.2808±0.3884	21.1475	0.9112
	N_{II}	6.4172±0.4008	25.9640	1.6424
	S	6.5011±0.3075	25.9004	2.8032
	A	6.4553±0.3185	22.1869	—
Males	N_I	6.0698±0.3678	14.4461	0.9017
	N_{II}	6.1239±0.3204	16.0262	1.6289
	S	6.1843±0.3200	15.8998	2.2265
	A	6.0045±0.4408	11.7328	—
mean exuvia		5.4125±0.0969		—

The caloric equivalent (E, cal/indiv. — Tab. VI), i.e. the caloricity of a mean individual, increased for the successive stages and attained a maximum for nymphs N_{II} of both sexes. In the course of the development of the aquatic forms, the caloric equivalent (E, cal/indiv.) increased with a rise of dry weight (W_{dry}, mg), according to the equation:

$E=5.957\ W_{dry}{}^{1.046}$ (correlation coefficient $r=0.99$). On the other hand, after metamorphosis, in the winged forms which do not feed (subimago imago), the caloric equivalent decreased (by about 15 and about 27% in females and males, respectively), as compared with its value for nymphs N_{II}.

It was assumed that the caloricity of exuviae does not change during the development. The mean caloric value of exuviae of *C. dipterum* was 5.41 cal/mg dry weight. Moreover, the caloric equivalent of exuviae (E, cal/exuvia) was calculated for each stage (Tab. VI).

The ash content in the investigated material was estimated only from the residues after combustion in the microbomb; ash accounted, on the average, for about 4% of dry body weight of animals and for about 7% of dry weight of exuvia.

C. OXYGEN CONSUMPTION AT 20° C

For the youngest larval stages (L_I, L_{II}, L_{III}), 23 measurements of the oxygen consumption were performed in manometric respirometers (Cartesian divers) and 50 measurements — in volumetric respirometers. The

Table VII. Oxygen consumption of the youngest larval stages of *Cloëon dipterum* at 20°C—comparison of two methods

Methods of measurement:		Manometric respirometer	Volumetric respirometer	Stages
n		23	50	L_{I}—L_{III}
Wet weight (mg)	Means	0.431	0.428	
	min-mix	0.16−0.89	0.14−0.94	
Oxygen consumption—R (cm^3 O$_2$/h · indiv.)	$\bar{x}\pm$SD	0.224±0.057	0.219±0.018	L_{I}
	t	0.03−NS		
	$\bar{x}\pm$SD	0.228±0.042	0.252±0.047	L_{II}
	t	0.52−NS		
	$\bar{x}\pm$SD	0.344±0.075	0.400±0.114	L_{III}
	t	0.53−NS		
	$\bar{x}\pm$SD	0.267±0.080	0.315±0.115	L_{I}—L_{III}
	t	0.46−NS		

values obtained with the use of divers were on the whole somewhat lower (NS) (Tab. VII); since the results obtained by the two methods did not differ significantly, they were considered jointly.

The volumes of oxygen (R, mm^3 O$_2$/h) consumed at 20°C by a mean individual in the successive developmental stages were compared (Tab. VIII, Fig. 8), and significant differences (HS) between the individual

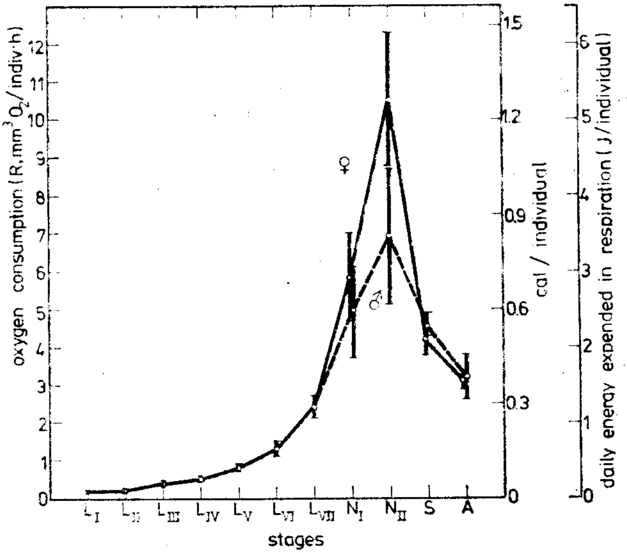

Fig. 8. Oxygen consumption (R, mm^3 O$_2$/h·indiv.) at 20°C in each developmental stage of *C. dipterum*, and 24-h energy expenditure for respiration (cal/indiv.); the values of $\bar{x}\pm$ci are given

Table VIII. Mean oxygen consumption in the succesive developmental stages of *C. dipterum* at 20°C; 1 cal = 4.184J

Stage		n	Wet weight (mg)		Oxygen consumption in mm³ O_2/h					cal/24 h
					per mg wet weight	per mg dry weight	per individual			
			min	max	$\bar{x}\pm SD$	$\bar{x}\pm SD$	$\bar{x}\pm SD$	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	p	$\bar{x}\pm SD$
L_I		15	0.14	0.38	1.123±0.249	5.067±1.645	0.190±0.033			0.0228±0.0040
								0.052±0.028	HS	
L_{II}		27	0.26	0.54	0.784±0.162	3.221±0.715	0.242±0.045			0.0290±0.0054
								0.153±0.046	HS	
L_{III}		31	0.37	0.94	0.668±0.151	2.790±0.669	0.395±0.112			0.0474±0.0134
								0.143±0.074	HS	
L_{IV}		20	0.53	1.52	0.496±0.116	2.053±0.395	0.538±0.143			0.0646±0.0172
								0.269±0.103	HS	
L_V		22	1.32	2.82	0.372±0.068	1.647±0.290	0.807±0.177			0.0968±0.0212
								0.512±0.238	HS	
L_{VI}		22	1.93	4.97	0.302±0.074	1.302±0.250	1.319±0.512			0.1583±0.0614
								1.077±0.386	HS	
L_{VII}		24	2.61	7.80	0.334±0.064	1.489±0.248	2.396±0.731			0.2875±0.0877
								3.471±0.786	HS	
Females	N_I	8	7.04	16.00	0.468±0.096	1.841±0.479	5.867±1.314			0.7040±0.1577
								4.640±2.118	HS	
	N_{II}	9	15.39	21.24	0.648±0.130	2.590±0.629	10.507±2.333			1.2608±0.2800
								−6.292±3.665	HS	
	S	10	12.58	16.99	0.131±0.020	1.058±0.062	4.215±0.608			0.5058±0.0730
								−1.074±0.475	HS	
	A	11	7.48	12.19	0.328±0.069	0.914±0.099	3.141±0.353			0.3769±0.0424
								2.551±1.151	HS	
Males	N_I	6	4.21	10.17	0.593±0.137	1.945±0.529	4.947±1.159			0.5936±0.1391
								t=1.76	NS	
	N_{II}	7	8.83	12.29	0.774±0.136	2.645±0.397	6.922±1.945			0.8306±0.2334
								−2.348±1.408	HS	
	S	10	7.22	11.58	0.591±0.100	1.779±0.241	4.574±0.398			0.5489±0.0478
								−1.338±0.615	HS	
	A	10	3.67	5.40	0.681±0.128	1.656±0.277	3.236±0.829			0.3883±0.0995

stages were found. Only the differences in respiration between nymphs N_I and N_{II} of males were insignificant (NS). In all stages (Tab. IX, Fig. 8), the females consumed more oxygen than males, but the differences were significant (HS) only for stage N_{II}.

Oxygen consumption at 20°C was maximal for the stage of nymphs (N_{II}). With the use of the right scale in Fig. 8, readings were made of the 24-h energy losses for respiration (Tab. VIII), remaining within the range: from about 0.02 cal/indiv. for stage L_I to about 1.26 cal/indiv. for stage N_{II} in females, i.e. about 0.08 and 5.27 J/indiv., respectively.

During the development of *C. dipterum*, changes in the oxygen consumption rate ($mm^3\,O_2/h$ — calculated per unit of wet and dry weight) were observed (Tab. VIII, Fig. 9). At 20°C, a high oxygen consumption rate was shown by the stages of young larvae (maximum for stage L_I) and by nymphs before metamorphosis. A low oxygen consumption rate was found for the stages of big larvae (minimum for stage L_{VI}) and for the winged forms (subimago) of both sexes. Males after metamorphosis exhibited (Tab. IX, Fig. 9) a much higher (HS) oxygen consumption rate than females, whereas for the stages of nymphs no differences were found (NS).

Relationships between the oxygen consumption (R, $mm^3\,O_2/h \cdot$indiv.) and body weight (W_{wt}, mg and W_{dry}, mg) as well as between the linear dimensions of the body (l, mm and h, mm) were described by regression equations. Statistical comparison was made (Tabs. X and XI) of coefficients b of the calculated regressions for the successive stages. Common regressions were calculated for the developmental periods for which no differences (NS) between the successive stages were found (Tab. XII). All regressions were highly significant.

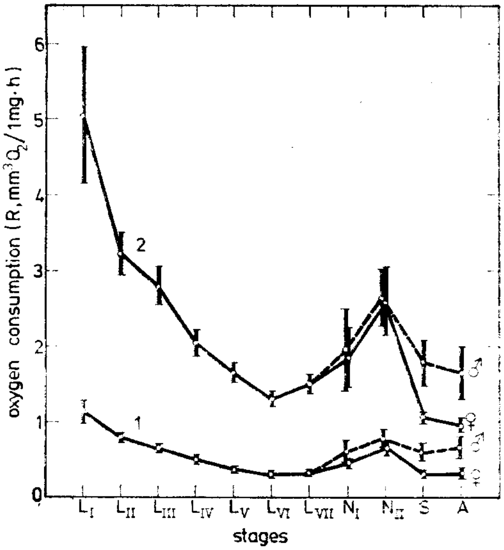

Fig. 9. Oxygen consumption rate ($\bar{x} \pm$ ci) at 20°C in the successive developmental stages of *C. dipterum*: 1 — oxygen consumption, $mm^3\,O_2/h \cdot mg$ wet weight, 2 — oxygen consumption, $mm^3\,O_2/h \cdot mg$ dry weight

Table IX. Mean oxygen consumption (at 20°C) as a function of sex in *Cloëon dipterum*

Stage	$mm^3\ O_2/h$:	per mg wet weight		per mg dry weight		per individual	
	Sex:	♀♀	♂♂	♀♀	♂♂	♀♀	♂♂
N_I	$\bar{x}\pm$SD	0.468 ± 0.096	0.593 ± 0.137	1.841 ± 0.479	1.945 ± 0.529	5.867 ± 1.314	4.947 ± 1.915
	t	0.30 – NS		0.38 – NS		1.36 – NS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	—		—		—	
N_{II}	$\bar{x}\pm$SD	0.648 ± 0.130	0.774 ± 0.136	2.590 ± 0.629	2.645 ± 0.397	10.507 ± 2.333	6.922 ± 1.945
	t	1.88 – NS		0.20 – NS		3.27 – HS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	—		—		3.585 ± 2.510	
S	$\bar{x}\pm$SD	0.313 ± 0.020	0.591 ± 0.100	1.058 ± 0.062	1.779 ± 0.241	4.215 ± 0.608	4.574 ± 0.398
	t	8.62 – HS		9.16 – HS		1.56 – NS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	0.278 ± 0.071		0.721 ± 0.174		—	
A	$\bar{x}\pm$SD	0.328 ± 0.069	0.681 ± 0.128	0.914 ± 0.099	1.656 ± 0.277	3.141 ± 0.353	3.236 ± 0.829
	t	7.97 – HS		8.34 – HS		0.35 – NS	
	$(\bar{x}_1-\bar{x}_2)\pm L_{0.05}$	0.353 ± 0.098		0.741 ± 0.196		—	

Table X. Comparison of coefficients b of regressions of the relationship between oxygen consumption (R, mm³ O_2/h · individual) and body weights (W_{wt}, mg and W_{dry}, mg) in some selected developmental stages of *Cloëon dipterum*—at 20°C. The numbers represent the t value (acc. to Simpson et al. 1960); HS—highly significant differences, S—slightly significant differences, NS—no differences

Stages	L_{I}	L_{II}	L_{III}	L_{IV}	L_{V}	L_{VI}	L_{VII}	N♀♀	N♂♂
L_{I}	XX	0.72 NS	2.01 NS	1.38 NS	1.02 NS	3.70 HS	4.89 HS	2.92 HS	5.20 HS
L_{II}	0.90 NS	XX	0.89 NS	0.35 NS	0.20 NS	2.62 HS	2.64 HS	2.12 HS	4.14 HS
L_{III}	1.86 NS	1.05 NS	XX	0.63 NS	0.82 NS	1.97 S	1.99 S	1.46 NS	3.46 HS
L_{IV}	2.19 S	1.16 NS	0.06 NS	XX	0.22 NS	2.52 HS	2.63 HS	1.99 S	4.08 HS
L_{V}	1.68 NS	0.57 NS	0.55 NS	0.65 NS	XX	2.64 HS	2.75 HS	2.11 S	4.31 HS
L_{VI}	3.08 HS	2.19 S	1.41 NS	1.42 NS	1.96 S	XX	0.69 NS	0.43 NS	0.90 NS
L_{VII}	4.23 HS	.230 S	1.08 NS	0.62 NS	1.99 S	0.85 NS	XX	0.16 NS	2.07 S
N♀♀	2.44 S	1.63 NS	0.88 NS	0.86 NS	1.29 NS	0.39 NS	0.29 NS	XX	1.32 NS
N♂♂	3.55 HS	2.43 S	1.59 NS	1.97 S	2.11 S	0.14 NS	1.09 NS	0.54 NS	XX

Relationship: R (mm³ O_2/h · indiv.) and W_{dry} (mg)

Relationship: R (mm³ O_2/h · indiv.) and W_{dry} (mg)

It was found (Tab. XI) that only the relationship between the oxygen consumption and body length (R/l) was constant in the development of larvae and nymphs. On the grounds of 191 measurements of the oxygen consumption of individuals from stages L_I—N_{II} of *C. dipterum* (body length 2.29—10.29 mm), the equation of exponential regression was calculated (Tab. XII). This equation, in a logarithmic form and upon application of a 95% confidence interval for coefficients a and b of the regression, assumes the form:

$$\log R = (2.6027 \pm 0.0525) + 0.4343\,(0.5844 \pm 0.0222) \cdot l,$$

where: $0.4343 = \log e$. Also a graphic interpretation of this equation (Fig. 10) confirmed the statistical calculations: on a semilogarithmic scale, the points representing the measurements were arranged along one straight line.

An analysis of the remaining relationships (Tabs. X and XI) showed that in the development of the aquatic forms of *C. dipterum* two periods differing in the oxygen consumption rate could be distinguished (Tab. XII). Points representing the measurements of respiration for the second period were arranged along steeper lines (Figs. 11 and 12). Oxygen consumption of older larvae and nymphs was higher than it could be expected from their body weights. The relationship between the oxygen

Table XI. Comparison of coefficients *b* of regression of the relationship between oxygen consumption (R, mm^3 O$_2$/h · individual) and linear body dimensions (l, mm and h, mm) in some selected developmental stages of *Cloëon dipterum*—at 20° C. Denotations as in Tab. X

Stages	L_I	L_{II}	L_{III}	L_{IV}	L_V	L_{VI}	L_{VII}	N♀♀	N♂♂
L_I	XX	0.55 NS	0.59 NS	1.18 NS	0.58 NS	0.17 NS	1.19 NS	0.20 NS	0.11 NS
L_{II}	0.45 NS	XX	0.01 NS	0.50 NS	0.09 NS	0.47 NS	0.36 NS	0.83 NS	0.38 NS
L_{III}	0.76 NS	0.48 NS	XX	0.55 NS	0.10 NS	0.52 NS	0.41 NS	0.89 NS	0.40 NS
L_{IV}	1.33 NS	1.09 NS	0.50 NS	XX	0.75 NS	1.23 NS	0.30 NS	1.53 NS	0.86 NS
L_V	0.61 NS	0.19 NS	0.36 NS	0.98 NS	XX	0.51 NS	0.71 NS	0.91 NS	0.36 NS
L_{VI}	3.50 HS	3.42 HS	2.04 S	1.41 NS	3.27 HS	XX	1.37 NS	0.51 NS	0.01 NS
L_{VII}	3.94 HS	4.02 HS	2.33 HS	1.32 NS	3.81 HS	0.32 NS	XX	1.64 NS	0.89 NS
N♀♀	0.98 NS	0.80 NS	0.41 NS	0.01 NS	0.72 NS	1.07 NS	0.95 NS	XX	2.48 S
N♂♂	1.80 NS	1.62 NS	1.07 NS	0.63 NS	1.52 NS	0.46 NS	0.31 NS	0.53 NS	XX

Relationship: R (mm^3 O$_2$/h · individual) and h (mm)

Relationship: R (mm^3 O$_2$/h · individual) and l (mm)

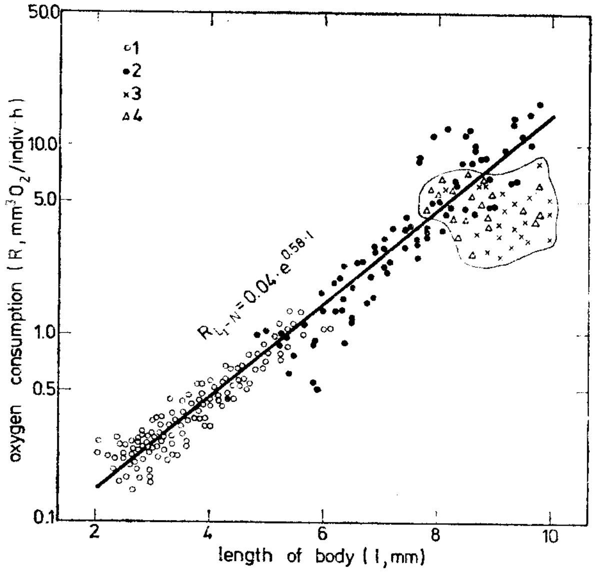

Fig. 10. Relationship between oxygen consumption (R, mm^3/h·indiv.) and body length (l, mm) in *C. dipterum* at 20°C (semi-logarithmic scale): 1 — stages L_I—L_V (n=115), 2 — stages L_{VI}—N (n=76), 3 — females, stages S and A (n=21), 4 — males, stages S and A (n=20)

Table XII. Relationships between oxygen consumption (R, mm^3 O$_2$/h · indiv.) and body weight (W_{wt}, mg and W_{dry}, mg), and linear dimensions (l, mm and h, mm) in *C. dipterum*—at 20°C

Relationship calculated		Stages	n	Dispersion range (min–max)	Regression equations (with 95% confidence interval for coefficient b)	r
Oxygen consumption—R (mm^3 O$_2$/h · individual)	R/W_{wt}	L_I–L_V	115	0.14– 2.82 mg	$R=0.4794 \cdot W_{wt}{}^{0.5624\pm0.0488}$	0.9066
		L_{VI}–N	76	1.93–21.24 mg	$R=0.1741 \cdot W_{wt}{}^{1.4658\pm0.0955}$	0.9633
		♀♀(S+A)	21	7.48–15.87 mg	$R=0.6722 \cdot W_{wt}{}^{0.6989\pm0.2479}$	0.8840
		♂♂(S+A)	20	3.67–11.58 mg	$R=1.0000 \cdot W_{wt}{}^{0.6904\pm0.2255}$	0.8586
	R/W_{dry}	L_I–L_V	115	0.02– 0.68 mg	$R=1.0789 \cdot W_{dry}{}^{0.5605\pm0.0505}$	0.8999
		L_{VI}–N	76	0.42– 5.40 mg	$R=1.4703 \cdot W_{dry}{}^{1.3058\pm0.0843}$	0.9635
		♀♀(S+A)	21	3.15– 4.64 mg	$R=0.7270 \cdot W_{dry}{}^{1.2132\pm0.6870}$	0.7987
		♂♂(S+A)	20	1.79– 3.25 mg	$R=1.4890 \cdot W_{dry}{}^{1.1088\pm0.6344}$	0.8185
	R/l	L_I–N	191	2.29–10.29 mm	$R=0.0401 \cdot e^{0.5844\pm0.0222l}$	0.9667
	R/h	L_I–L_V	115	0.32– 1.05 mm	$R=0.0717 \cdot e^{2.4619\pm0.2725h}$	0.8573
		L_{VI}–N	76	0.93– 1.53 mm	$R=0.0055 \cdot e^{5.0154\pm0.3879h}$	0.9489

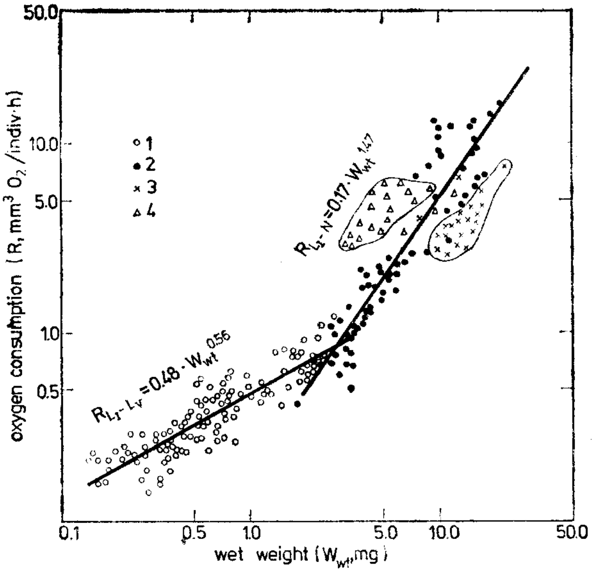

Fig. 11. Relationship between oxygen consumption (at 20°C) and wet weight (W_{wt}, mg) in *C. dipterum* (logarithmic scale); denotations 1, 2, 3 and 4 — as in Fig. 10

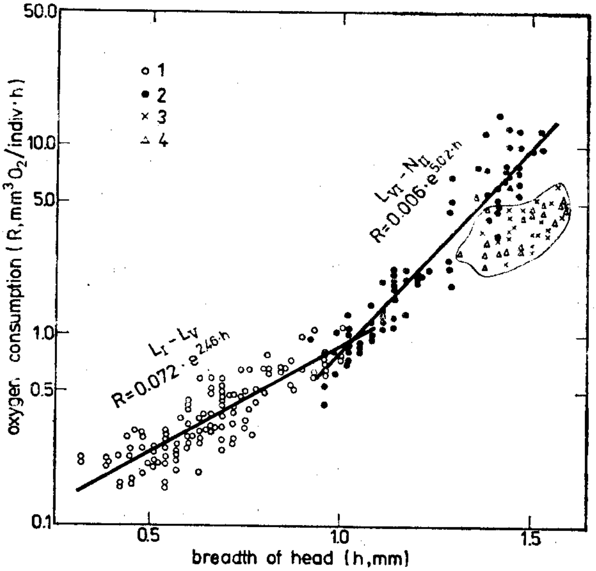

Fig. 12. Relationship between oxygen consumption (at 20°C) and head capsule breadth (*h*, mm) in *C. dipterum* (semi-logarithmic scale); denotations 1, 2, 3 and 4 — as in Fig. 10

consumption (R, mm^3 O$_2$/h·indiv.) and wet body weight (W_{wt}, mg) for the above-mentioned developmental periods was described by logarithmic regression equations (Tab. XII). Below we present the logarithmic form of these equtions (with application of 95% confidence intervals for coefficients a and b of the regresions).

For stages from L$_{\text{I}}$ to L$_{\text{V}}$ (n=115):

$$\log R=(1.6810\pm0.0191)+(0.5624\pm0.0488)\cdot\log W_{wt};$$

and for stages from L$_{\text{VI}}$ to N$_{\text{II}}$ (n=76):

$$\log R= (1.2406\pm0.0794)+(1.4658\pm0.0955)\cdot\log W_{wt}.$$

The big differences in oxygen consumption between the two distinguished developmental periods, evident from the distribution of points in the plot (Fig. 11), were confirmed also by statistical tests (d_b=37.64 — HS).

For the winged forms, the relationships: R/W_{wt} (Tab. XII, Fig. 11) and R/W_{dry} (Tab. XI) were described, separately for each sex, by logarithmic regression equations. No correlation between the volume of oxygen consumed and linear dimensions was found for these stages (correlation coefficient r was nearly zero).

The change in the oxygen requirement during the development of *C. dipterum*, expressed by an increase in the oxygen consumption rate and higher coefficients b of regressions of R/W_{wt}, R/W_{dry} and R/h before metamorphosis, indicates that possibly in the older larvae and nymphs the physiological processes are more intense that in the younger developmental forms.

4. DISCUSSION

As expected (Winberg 1968), the curves of the increase in body weights of *C. dipterum* are of a typically S-shaped nature (Fig. 3). Changes in the linear dimensions of the body, similarly as in the other mayflies studied from this angle (e.g. Trama 1957; Bretschko 1965; Clifford 1970), are relatively uniform and proportional to the stage (Fig. 2).

The equation describing the relationship between wet and dry body weight of *C. dipterum*, as calculated in this study, differs significantly from an analogous equation given by Pattée (1968): $W_{wt}=0.1+4.6\,W_{dry}$ (with the use of significance tests we obtained: $d_b=2.48$ — HS). However, it is stressed that the equation given by Pattée results from studies of animals whose dispersion range of weights (1.6—5.3 mg wet weight) is too narrow. Individual variability greatly influences the fluctuations of the slope of the regression lines, this resulting — in case of a small dispersion of body weights — in a wide 95% confidence interval of coefficient *b* (Pattée has obtained a value of *b* remaining between 4.1—5.1). An additional Pattée's error consists of the use, in the calculations, of mean body weight obtained by weighing of 44 groups, 2—30 individuals each. Moreover, drying at 110° C (Pattée) might have caused losses of a part of organic matter and underestimation of dry body weight (Dowgiałło 1975). In the present studies, the greater number of measurements ($n=1096$) involving animals with wide dispersion of body weights (0.05—21.24 mg wet weigth) permitted us to calculate a regression equation with a more narrow 95% confidence interval for coefficient *b* (from 3.8 to 4.1). Thus, this equation was applied for conversion of wet and dry weight of *C. dipterum*, though it is not necessarily of universal applicability. It seems that the water content (79%) in the body of larvae and nymphs of *C. dipterum*, as reported by Pattée (1968), is also somewhat overestimated; our results (Tab. III) are by about 1—8% lower.

Considerations of the regressions between the biometric parameters (Tab. V) showed that the rate of the increase in dry weight relative to body length (Fig. 6) is nearly constant, and that it is described by one regression equation for the whole developmental period of larvae and nymphs of *C. dipterum*. On the other hand, the increase in wet weight relative to the linear dimensions (Fig. 7) is more rapid for the young larval stages than for the later developmental stages. The change in the rate of the increase in wet weight on the turn of stages L_V and L_{VI} is related to a drop in body hydration in older larvae (Fig. 5).

It is known (Winberg 1968) that in case of isometric growth (without changes in the shape of the body) coefficient *b* of the regression of the relationship between body weight and the linear dimensions approaches 3. Values of coefficients *b* of the regression obtained for *C. dipterum* (Tab. V) testify to "gaining weight" in the aquatic stages, whereas in the winged stages these animals "lose weight".

The development of *C. dipterum* is associated with an increase in the caloric value of the body and with an increase in the caloric equivalents in the successive stages. The values obtained (Tab. VI) correspond to the mean caloricity of aquatic invertebrates, amounting to 4.42—6.64 kcal/g dry weight (Prus 1970). The caloric value was maximal for the subimago stage (by about 29 and about 23% higher in females and males, respectively, as compared with stage L_I). Similar values have been obtained by Trama (1957) for another species of Ephemeroptera — *Stenonema pulchellum*; this author has likewise observed an increase in body caloricity during the development of this species (5.290 and 5.710 cal/mg for nymphs 4 and 7 mm of length, respectively).

The oxygen consumption values were also consistent with the literature data. Fox, Simmonds (1933) have found for *C. dipterum* larvae a mean oxygen consumption of 1.780 $cm^3 O_2/h \cdot g$ dry weight (at 17°C). Kamler (1970) has reported for *C. dipterum* larvae with mean dry weight of 0.21 mg an oxygen consumption amounting to 1.859—2.655 cm^3 $O_2/h \cdot g$ dry weight (at 20°C). In the present studies a mean individual of the aquatic stages (L_I—N), weighing 0.36 mg dry weight, consumed 2.17 $cm^3 O_2/h \cdot g$ dry weight. Trama (1957) has found for *Stenonema pulchellum* a mean value of 1.74 cm^3 $O_2/h \cdot g$ dry weight (at 20°C).

Respiration measurements were performed at 20°C, this temperature remaining within the optimal range for the investigated species (Ivanova 1958). Moreover, at this temperature the oxygen consumption is high enough to permit detection of the more distinct differences between the investigated stages. The size of the respirometric vessels enabled relative motor activity, but during the measurements the animals were deprived of food. However, Kamler (1969) has observed that even 50-h starving fails to influence considerably the respiration of *C. dipterum*. Ivanova (1958) has reported that in larvae of this species digestion causes an increase in the metabolism by only 8.4—13.4% (mean about 11%). Studying separately the respiration of immobile, active, starved and fed larvae, Ivanova (1958) has calculated their 24-h oxygen consumption; a *C. dipterum* larva weighing 5.7 mg wet weight consumed an average of about 8.65 cm^3 O_2/g wet weight per 24 h, this representing a "cost"=41.74 cal/g wet weight per 24 h (24-h losses of caloricity for respiration amounted to about 0.24 cal/indiv.). Calculation of our own data (Fig. 8) afforded values approaching the results of Ivanova (1958), i.s. a "cost"=46.32 cal/g wet weight per 24 h, i.e. about 0.27 cal/indiv. (for body weight amounting also to 5.7 mg wet weight).

Trama (1957) has calculated for *Stenonema pulchellum* nymphs (4—6 mm long) a mean maintenance cost of about 0.24 cal/indiv. per 24 h (a mean individual required 8.07 cal for an increase in body length by 1 mm during 33 days).

Changes in the oxygen consumption rate during the development of *C. dipterum*, as observed in these studies (Fig. 9), seem to be characteristic of Ephemeroptera. Likewise, Ivanova (1958) has observed a high oxygen consumption rate in the youngest forms of *C. dipterum* (about 0.83 $cm^3 O_2/h \cdot g$ wet weight) and in the oldest forms, whereas for medium larvae (about 5 mg) a minimum value (about 0.22 $cm^3 O/_2h \cdot g$ wet weight) has been obtained. Hilmy (1962) has found similar fluctuations in the oxygen consumption rate during the development of three other species of Ephemeroptera (*Habroleptoides modesta, Hobroleptophlebia lauta* and *Ephemerella ignita*).

The course of changes (Fig. 13) in oxygen consumption ($cm^3 O_2/h$), calculated per unit of weight (curve 1 — per mg dry weight. Curve 2 — per mg wet weight) and per one individual (curve 3), as a function of changes in wet body weight (W_{wt}, mg) in the successive development stages of *C. dipterum* indicates that the nature of curves 1, 2 and 3 is

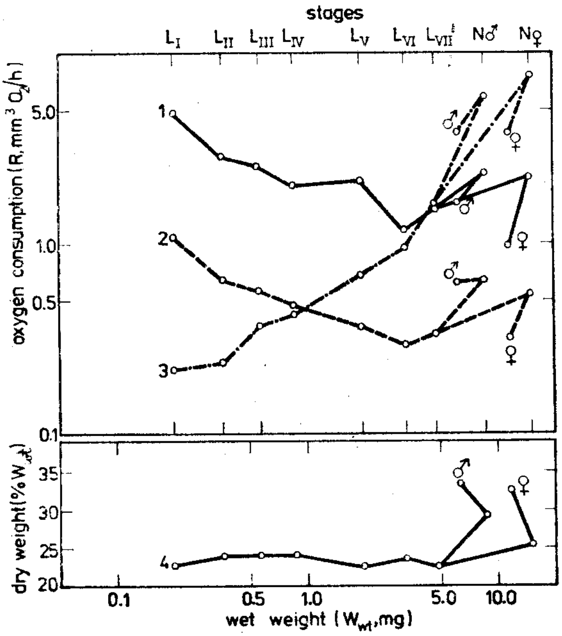

Fig. 13. Oxygen consumption (at 20°C) in the successive developmental stages of *C. dipterum* as a function of wet weight: 1 — oxygen consumption, mm^3 O$_2$/h·1 mg dry weight, 2 — oxygen consumption, mm^3 O$_2$/h·1 mg dry weight, 3 — oxygen consumption, mm^3 O$_2$/h·indiv., 4 — percentile content of dry weight in bodies of larvae and nymphs

consistent with the general biological regularities. At the successive stages, oxygen consumption of a mean individual (curve 3) rises with age, because the body biomass increases. The drop for the winged stages (drop of curves 1, 2 and 3) is caused by a decrease in body weight and by the economical energy metabolism of the non-feeding forms. Curves illustrating the oxygen consumption rate (1 and 2) at first proceed in parallel; the differences observed for the final stages of development are related to a drop in body hydration, which is abrupt at this time (curve 4). From the stage of nymphs, sexual differentiation is reflected by the respiration, since the oxygen consumption rate indices are higher for males. In the period from stage L$_{VI}$ to transformation of nymphs, curves 1, 2 and 3 indicate an increase in oxygen consumption which is faster than it could be expected from body weights. On the turn of stages L$_V$ and L$_{VI}$, the oxygen consumption rate increased (Tab. VIII, Fig. 9); coefficients *b* of the regression of the relationship between the oxygen consumption rate and body weight were also higher (Tab. XII, Figs. 11 and 12).

In classical regression (Winberg 1956), the relationship between the oxygen consumption rate (R, mm^3 O$_2$/h·indiv.) and body weight (W_{wt}, mg), calculated for two species of genus Cloëon (body weights 0.19—10.25 mg), is expressed by the equation:

$$R=0.872\,W_{wt}^{0.88}$$

Alimov (1971) and Ivanova, Alimov (1973) have reported, consistently with Winberg (1956), for Ephemeroptera larvae (weighing 0.6—151 mg) the following general equation:

$$R = 1.435\ W_{wt}^{0.803}$$

In the present studies we calculated one common regression from all oxygen consumption measurements ($n = 191$) by individuals of the aquatic stages of *C. dipterum*, and obtained:

$$R = 0.548\ W_{wt}^{(0.835 \pm 0.040)}$$

In the above equations of the Russian authors, the high value of intercept a, being about twice higher than that calculated from our own results, is noteworthy. It can be assumed that the above authors have calculated the regression from the results of oxygen consumption measurements made for only two groups of larvae: the youngest and oldest ones. We calculated the equation of regression from our own measurements obtained for the two above groups of larvae, without consideration of the intermediate stages, and found for intercept a the value of 0.86, being closer to that reported by Winberg 1956 and Alimov 1971.

The value of coefficient b of the regression calculated for all investigated developmental stages of *C. dipterum* is closely similar to the value reported by the above authors. However, statistical comparison of coefficient b of this regression (0.835) with coefficients b of the regressions calculated separately for the two developmental periods (Tab. XII, Fig. 11) gives $d_b = 8.62$ (for period I_I—L_V) and $d_b = 12.14$ (for period L_{VI}—N). Thus, the relationship between oxygen consumption and body weight of *C. dipterum* cannot be described by the common regression equation, because this regression differs significantly (HS) from the regressions calculated separately for both age groups and it does not correspond to the real relationship R/W_{wt} for the developmental periods of this species. Therefore, for ecological and bioenergetic purpose it seems justified to use two regressions in the calculations, consistently with Tab. XII.

An analysis of the biometric data suggests the occurrence, in the development of *C. dipterum*, of two periods differing in the intensity of the physiological processes. Results of oxygen consumption measurements confirm this assumption. The initial period of larval development (stages I_I—L_V) is characterized by a rapid rate of increase in body weight (see chapter: 3A) which is probably equal for both sexes. In laboratory cultures (**Cianciara** 1980), at stage L_V the first differences between larvae of both sexes occurred, since the females began to predominate over males in body length (i.e. in body weight as well). In the second period of the development of *C. dipterum* (stages L_{VI}—N), the increase in wet weight relative to the body dimensions is somewhat slower, whereas the metabolic processes become more intense, as expressed by a much higher oxygen consumption. The high oxygen consumption rate, found for the second developmental period, is probably related to the abrupt development of gonads at this time (Soldan, pers. comm.). The most intense development and growth of the sexual glands in females takes place earlier, probably at the stages of older larvae (L_{VI}, L_{VII}); this intensifies the physiological processes in a mean individual of this species during the period before in these studies sex has been distinguished. The higher oxygen consumption rate in nymphs and winged forms of males may be due to the later maturation of gonads in this sex.

5. SUMMARY

The present studies comprised the determination of the biometric parameters (body dimensions and weights) and bioenergetic parameters (caloric value, respiration) for the successive developmental stages of *C. dipterum*: larvae (stages L_{I}—L_{VII}), nymphs (N_{I}, N_{II}), subimago (S) and imago (A). A total of 1149 measurements of body length, head capsule breadth, and wet and dry body weight, as well as 89 combustions in a calorimetric microbomb and 232 measurements of oxygen consumption at 20°C were performed. The course of changes in the measured values with the development of *C. dipterum* was analyzed. It was found that the linear dimensions increase uniformly and in proportion to the size of the stage; the curves of the increase in wet and dry weight are S-shaped. The caloric value of the body increases with the development from 5.02 cal/mg dry weight (for stage L_{I}) and attains a maximum at the subimago stage (6.50 and 6.18 cal/mg dry weight in females and males respectively). At the successive stages, oxygen consumption of a mean individual increases with age and with a rise of body biomass, whereas the oxygen consumption rate varies, being highest for stage L_{I} and lowest for stage L_{VI} (5.07 and 1.30 $mm^3\,O_2/h \cdot mg$ dry weight, respectively). An analysis of the biometric data showed that the rate of the increase in dry weight is constant during the development of larvae and nymphs of *C. dipterum*, and that it is expressed by the equation: $W_{dry}=0.001 \cdot l^{3.68}$ ($r=0.98$), whereas the increase in wet weight relative to body length is faster in young larvae (stages L_{I}—L_{V}) than in the later development (stages L_{VI}—N). On the other hand, the younger stages exhibited a much lower oxygen consumption ($R=0.48\, W_{wt}^{0.56}$) relative to body weight than the older forms ($R=0.17 \cdot W_{wt}^{1.47}$).

Thus, the development of *C. dipterum*, while characterized by a constant rate of the increase in dry weight relative to body length and by a constant increase in the caloric value of the body, exhibits two periods differing in the intensity of the physiological processes: period I (comprising stages L_{I}—L_{V}) characterized by a higher rate of the increase in wet weight (and a rise of body hydration), and period II (comprising stages L_{VI}—N) characterized by higher oxygen consumption (and a drop in body hydration).

6. STRESZCZENIE

W przeprowadzonych badaniach wyznaczono wartości parametrów biometrycznych (rozmiary i ciężar ciała) i bioenergetycznych (wartość kaloryczna, respiracja) w kolejnych stadiach rozwoju *C. dipterum*: larw (stadia L_{I}—L_{VII}), nimf (N_{I}, N_{II}), subimago (S) i imago (A). Ogółem wykonano 1149 pomiarów długości ciała, szerokości puszki głowowej, mokrej i suchej masy, 89 spaleń w mikrobombie kalorymetrycznej oraz 232 pomiary zużycia tlenu w 20°C. Analizowano przebieg zmian zmierzonych wartości w miarę rozwoju *C. dipterum*. Stwierdzono, że wymiary liniowe wzrastają równomiernie i proporcjonalnie do wzrostu stadium, zaś krzywe wzrostu mokrej i suchej masy są S-kształtne. Wartość kaloryczna ciała wzrasta wraz z rozwojem od 5,02 cal/mg suchej masy (w stadium L_{I}) i osiąga maksimum w stadium subimago: 6,50 cal/mg suchej masy (samice) i 6,18 cal/mg suchej masy (samce). Zużycie tlenu przez przeciętnego osobnika na danym etapie rozwoju wzrasta wraz z wiekiem i wzrostem biomasy ciała, lecz intensywność zużycia tlenu jest zmienna: najwyższa w stadium L_{I} (5,07 $mm^3\,O_2$/godz. mg suchej masy), a najniższa w stadium L_{VI} (1,30 mm^3

O_2/godz. mg suchej masy). Analizując dane biometryczne stwierdzono, że tempo wzrostu suchej masy jest stałe w rozwoju larw i nimf *C. dipterum* i wyrażone równaniem: $W_{dry}=0{,}001 \cdot l^{3.68}$ ($r=0{,}98$), podczas gdy wzrost mokrej masy w stosunku do długości ciała u młodych larw (stadia L_{I}—L_{V}) jest szybszy, niż w późniejszym okresie rozwoju (stadia L_{VI}—N). W młodszych stadiach stwierdzono natomiast znacznie niższe zużycie tlenu ($R=0{,}48 \cdot W_{wt}^{0{,}56}$) w stosunku do masy ciała, niż u form starszych ($R=0{,}17 \cdot W_{wt}^{1{,}47}$).

Tak więc, w rozwoju *C. dipterum* przy stałym tempie wzrostu suchej masy w stosunku do długości ciała oraz stałym wzroście wartości kalorycznej ciała istnieją dwa etapy o różnym nasileniu procesów fizjologicznych: etap I (obejmujący stadia L_{I}—L_{V}) charakteryzujący się szybszym tempem przyrostu mokrej masy (i wzrostem hydratacji ciała) oraz etap II, obejmujący stadia L_{VI}—N — wyższe zużycie tlenu (i spadek uwodnienia ciała).

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